PB-227 645

# BIO-ASSAY INVESTIGATIONS, HOOKER ELECTROCHEMICAL COMPANY, NIAGARA FALLS, NEW YORK

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Robert A. Taft Sanitary Engineering Center
Cincinnati, Ohio

1956

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# BIO-ASSAY INVESTIGATIONS

# FOR

# INTERNATIONAL JOINT COMMISSION

HOOKER ELECTROCHEMICAL COMPANY

Niagara Falls, New York

1956

U. S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
Public Health Service
Robert A. Taft Sanitary Engineering Center
Cincinnati, Ohio

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# Bio-Assay Investigations

Hooker Electrochemical Company

Niagara Falls, New York

Prepared By

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Sanitary Engineering Center, USPHS

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This report covers an investigation of the toxicity to fish of some of the principal effluents from the Hooker Electrochemical Company.

Chemical wastes may contain compounds which are toxic to aquatic life. These compounds when mixed with or under the influence of other non-toxic components of the effluent or receiving water may exert an entirely different toxicity from that of the pure compounds. Bio-assays were made to evaluate directly the toxicity of these chemically complex wastes.

Bio-assays were also made on hexachlorocyclopentadiene, a product manufactured by the company.

## Organization of Study

Bio-assays were made on effluent samples collected during the period January to March, 1956. Hooker and Public Health Service personnel collaborated in collecting 8 or 24 hour composite samples and in making flow measurements in some of the plant sewers. The Public Health Service performed bioassays in laboratory space provided by the National Aniline Division, Allied Chemical and Dye Corporation in Buffalo. Analyses for phenols and cyanide were made in the I.J.C. unit laboratory.

Bio-Assay Methods

Bio-assays were made essentially by the method recommended by the Toxicity Subcommittee of the Federation of Sewage and Industrial Wastes Associations (Sewage and Industrial Wastes, Vol. 23, No. 11, 1380, Nov. 1951). This method consists of preparing various concentrations of effluent in a selected dilution water, adding the test fish and observing their reactions over a definite time period. For ease in the plotting and interpreting of results it is desirable to make up test solutions with the percentage of effluent in a logarithmic series as 10, 5.6, 3.2, 1.8, 1.0, etc.

As the effluents were of unknown toxicity, exploratory or small scale tests were made to determine the approximate toxic range. Test solutions were prepared over a wide range of concentration (e.g., 100, 10, and 1.0 per cent effluent). Two fish were added to 2 liters of each concentration in 6-1/2 inch diameter, 1 gallon widemouth glass bottles. Observations for relatively short time periods indicated test concentrations necessary for the full scale experiments.

In full scale bio-assays, 10 fish are normally used for each test concentration. Five fish are added to 10 liter duplicate samples in 5 gallon widemouth bottles, 10 inches in diameter. In these tests, however, duplicates were not always used. Some results are based on single samples containing 5 fish in 10 liters. The intermediate concentrations tested were dependent upon information obtained from the exploratory tests. For example, if fish were killed in concentrations above 10 per cent and not affected in concentrations of 1 per cent, intermediates were set up within this range (e.g. 10, 5.6, 3.2, 1.8, and 1.0 per cent concentration of effluent).

The dilution water used was raw Lake Erie water obtained at the Buffalo,

New York, water plant. This water was hauled into the laboratory, allowed to come to room temperature, and aerated vigorously for at least one hour to bring it to equilibrium with atmospheric gases. Characteristics of this water at time of use were as follows: Dissolved oxygen 7.6-8.2 ppm; pH 8.0-8.2; total alkalinity ($CaCO_3$) 94-100 ppm; total acidity ($CaCO_3$) 0-1 ppm; versenate hardness ($CaCO_3$) 120-135 ppm.

The test fish used were fathead minnows (Pimephales promelas), ranging in length from 2 to 2-1/2 inches and in weight from 1 to 1-1/2 grams. These fish were obtained in uniform lots from the Newtown, Ohio, Fish Hatchery and acclimated to laboratory conditions. This species is of intermediate tolerance to chemicals comparable to bass, sunfish, perch, and other warm water species, and will tolerate fairly low oxygen conditions (1-2 ppm).

The bio-assays were made at ordinary laboratory temperatures which were within the range from 22 to 25° C. and compare favorably with maximum summer water temperatures in this area.

The tests are designed so that generally no oxygenation or aeration is needed. Absorption of atmospheric oxygen by the exposed water surface is normally adequate for fish requirements during the test period. However, in some bio-assays, high oxygen demand effluents may cause oxygen depletion. When necessary, dissolved oxygen can be maintained by bubbling pure oxygen through the test solution by means of a suitable arrangement of valves and small tubing. The rate (60-180 bubbles per minute) is adjusted to maintain adequate oxygen for fish survival with minimum agitation in order to prevent the loss of volatile materials.

The oxygen demand of these effluents was such that it was not necessary to add oxygen during any of the bio-assays.

Physical and chemical determinations (temperature, dissolved oxygen, pH, alkalinity, and hardness) were made on each concentration initially, after fish mortality, or at the completion of the test. This was done primarily to detect low D.O. levels so they could be controlled and to differentiate between fish mortality due to oxygen deficiency or acidity and toxic properties.

Effluents were normally brought into the laboratory in the afternoon immediately following collection and exploratory tests were set up at once. Observations made the following morning furnished information on the toxicity of the wastes essential for setting up the full scale tests.

Fish reactions were observed over a 96 hour period. From the mortality in different concentrations, 24, 48, and 96 hour $TL_m$ (Median tolerance limit) values were obtained. The median tolerance limit is the concentration of effluent in dilution water that kills just 50 per cent of the test fish. These values were obtained by straight line graphical interpolation from points representing per cent survival of fish and log concentrations of effluents which bracketed the 50 per cent point.

The 96 hour $TL_m$ was used to compute the dilution ratio, which is the ratio of a unit volume of effluent to dilution water which produces a 50 per cent mortality of the test fish. This ratio times the effluent flow gives the dilution volume or the total flow or volume of receiving water required to reduce the toxicity of the effluent to where a 50 per cent mortality of the test fish is produced. The following formula may be used for directly computing this dilution volume:

$$\frac{100 - TL_m}{TL_m} \times \text{Effluent Flow} = \text{Dilution Volume}$$

The dilution volumes represent conditions which would cause direct injury to fish upon short time exposure and are a direct comparative measure of toxicity. Liberal application factors must be applied to these results for complete protection of aquatic life. These factors are based on the following major considerations:

(1) The bio-assay procedure measures 50 per cent mortality during a relatively short time period in non-renewed solutions. This must be related to concentrations having no effect during continuous long time exposure.

(2) Test data apply directly to the species of fish used. While fathead minnows are of intermediate tolerance and compare favorably with many warm water "game" fishes, some locally important species of fish and fish food organisms may be more sensitive. Bio-assays conducted on other industrial wastes with a locally important forage fish, the emerald shiner (Notropis atherinoides), indicated this species are approximately twice as sensitive as fathead minnows.

(3) Some effluents may vary in toxicity. A few samples may give an indication of toxicity but maximum conditions may be missed. In many situations, maximum conditions of toxicity are the limiting factor as far as aquatic life is concerned.

(4) Some conditions which may generally reduce but in some cases increase the magnitude of the application factor are the loss of volatiles, hydrolysis, oxidation, precipitation, and changes in water quality characteristics of the receiving water.

Based on the best available information, as much as 10 times the computed dilution volume may be necessary for protection of aquatic life in the receiving water.

## RESULTS

### Physical and Chemical Characteristics

A general description and some of the chemical characteristics of the effluent samples are shown in Table 1.

Dissolved oxygen values in the effluent samples were generally adequate for aquatic life support. While one sample of 47th Street Sewer and the Plant 4 Sewer effluents were initially low in oxygen, the bio-assay dilutions in which fish mortality occurred contained sufficient amounts. The oxygen demand of all effluents was apparently low. All bio-assay test concentrations maintained adequate oxygen for fish support during the 96 hour test period without the addition of an artificial supply. For adequate protection of aquatic life, dissolved oxygen concentrations should be maintained at 5 ppm or above in waters receiving toxic materials.

The 47th Street, West End and Plant 4 Sewers were strongly acid with pH values ranging from 1.8 to 2.5. While values within this range would be rapidly fatal to fish, the buffering capacity of the diluent water and toxic properties of the effluents were such that fish mortality in the bio-assay test concentrations could not be attributed to acidity except in the March 47th Street Sewer sample. Generally, pH values below 5 and above 9 may have a direct lethal effect on fish. Within these values, however, a change in pH may have an effect by increasing or decreasing the toxicity of various chemicals to fish. The toxicity of metals, cyanides, and sulfides may be increased by a decrease in pH while the toxicity of ammonia and some other materials is greater at higher pH values.

Phenols and cyanide in all effluents were at levels which would be ex-

TABLE 1 - PHYSICAL AND CHEMICAL DATA

HOOKER ELECTROCHEMICAL EFFLUENTS

| Source | Date 1956 | Description Color, odor, etc. | Dissolved Oxygen ppm | pH | Alkalinity ($CaCO_3$) ppm | Acidity ($CaCO_3$) ppm | Hardness ($CaCO_3$) ppm | Phenols ppm | Cyanide ppm |
|---|---|---|---|---|---|---|---|---|---|
| 47th St. Sewer 24 hr. composite | 1-20 | Grayish cast Sweetish solvent odor Some suspended material Clear when settled | 6.2 | 2.0 | 0 | 740 | 1000 | 0.20 | 0 |
| 47th St. Sewer 24 hr. composite | 2-17 | " | 0.4 | 2.1 | 0 | 1450 | 550 | 0.19 | 0 |
| Same as above neutralized with lime | 2-17 | No additional precipitate when neutralized | 3.0 | 8.0 | 70 | 2 | 2200 | - | - |
| 47th St. Sewer 24 hr. composite | 3-15 | Gray color Sour odor Turbid | 3.4 | 2.5 | 0 | 230 | 260 | 0.25 | .01 |
| Same as above neutralized with lime | 3-15 | Heavy precipitate when neutralized | 7.0 | 8.0 | 24 | 0 | 500 | - | - |
| Diversion Sewers 8 hr. composite | 2-21 | Grayish-tan color Very faint solvent odor Slightly turbid | 8.6 | 8.6 | 104 | 0 | 160 | 0.07 | 0 |
| West End Sewer 8 hr. composite | 2-21 | Light gray color Sweetish solvent odor Some suspended material | 9.2 | 2.1 | 0 | 1020 | 175 | 0.16 | 0 |

Table 1 (Cont'd) - PHYSICAL AND CHEMICAL DATA

HOOKER ELECTROCHEMICAL EFFLUENTS

| Source | Date 1956 | Description Color, odor, etc. | Dissolved Oxygen ppm | pH | Alkalinity ($CaCO_3$) ppm | Acidity ($CaCO_3$) ppm | Hardness ($CaCO_3$) ppm | Phenols ppm | Cyanide ppm |
|---|---|---|---|---|---|---|---|---|---|
| Same as above neutralized with lime | 2-21 | No additional precipitate | 9.0 | 7.2 | 64 | 12 | 1200 | - | - |
| Plant 4 Sewer (North St.) 8 hr. composite | 3-15 | Grayish-tan color Sweetish solvent color Slightly turbid | 0.2 | 1.8 | 0 | 520 | 300 | 0 | 0 |

pected to have no adverse effect on aquatic life.

No residual chlorine was found in any of the effluent samples. Chlorine may be toxic to fish in concentrations as low as 0.1 ppm.

The chemical or chemicals responsible for the toxicity in these effluents were not determined by the Sanitary Engineering Center during this study.

Toxicity of Effluents to Fish

A summary of bioassay results showing the comparative toxicity of the different effluent samples to fish is shown in Table 2. The data on which these results are based are shown in the Appendix at the end of this report. Twenty-four, 48, and 96 hour $TL_m$ values and dilution volumes were computed as described in the section on bioassay methods.

The 47th Street, West End, and Plant 4 sewer effluents were highly toxic to fish. The Plant 4 effluent was the most toxic but flow volume was small and thus the total contribution of toxicity (dilution volume) was lower than that from some of the other effluent samples.

The 47th Street sewer effluent while highly toxic in January and February was greatly reduced in toxicity in March. The toxicity in the January and February samples was not from acidity and neutralization had little effect in reducing the toxicity. The small amount of toxicity in the March sample, however, was primarily due to acidity. The company upon receiving a verbal report on the toxicity of the effluents following testing of the January and February samples, immediately instituted an investigation in the plant which led to the discovery of an unsuspected processing loss. This was corrected prior to the March sampling time.

TABLE 2 - SUMMARY OF BIO-ASSAY DATA

HOOKER ELECTROCHEMICAL EFFLUENTS

| Source | Date 1956 | Effluent Flow cfs | $TL_m$ (Median Tolerance Limit) (Percent Concentration) 24 hr. | 48 hr. | 96 hrs. | Dilution Volume cfs |
|---|---|---|---|---|---|---|
| 47th St. Sewer (24 hr. composite) | 1-20 | 10.0 | 3.2 | 2.6 | 2.5 | 390 |
| 47th St. Sewer (24 hr. composite) | 2-17 | 10.0 | 4.1 | 4.1 | 4.1 | 234 |
| Same as above (Neutralized) | 2-17 | 10.0 | 6.2 | 5.0 | 4.4 | 217 |
| 47th St. Sewer (24 hr. composite | 3-15 | 10.0 | 22 | 22 | 22 | 37 |
| Same as above (Neutralized) | 3-15 | 10.0 | >56 | >56 | >56 | - |
| Diversion Sewer 8 hr. composite) | 2-21 | 23.1 | 100% Surv. | 100% Surv. | 100% Surv. (1) | - |
| West End Sewer (8 hr. composite) | 2-21 | 4.8 | 7.5 | 7.5 | 7.5 | 59 |
| Same as above (Neutralized) | 2-21 | 4.8 | 16 | 13 | 13 | 32 |
| Plant 4 Sewer (8 hr. composite) | 3-15 | 1.2 | 2.2 | 1.5 | 1.5 | 79 |

(1) Percent survival of fish in undiluted effluent.

Some of the toxicity in the West End Sewer was apparently due to acidity as the toxicity was reduced by a factor of two when the effluent was neutralized.

No toxicity was found in the diversion sewer.

The dilution volume given for the effluent samples when multiplied by ten give the estimated receiving water flow necessary for protection of aquatic life.

## CONCLUSIONS

These limited bioassay studies have shown that some effluents from this plant were toxic to aquatic life.

The experimental work in cooperation with the plant resulted in their finding a source of toxic material entering the effluents. The company states that they have developed and instituted specific control tests which are routinely run to assure that these toxic materials do not enter the effluent streams.

To evaluate completely the contribution of toxicity to the Niagara River by this plant, a thorough bioassay investigation of all major effluents would be necessary.

## TOXICITY OF HEXACHLOROCYCLOPENTADIENE

Bioassays were made on the compound hexachlorocyclopentadiene (C-56) furnished by the Hooker Electrochemical Company. This material is a highly reactive chlorocarbon used in the manufacture of insecticides and other end products.

Bioassays were made by previously described methods in the Cincinnati laboratories of the Robert A. Taft Sanitary Engineering Center. The tests were conducted in a constant temperature room at 22° C. Two different dilu-

tion waters, which encompass the pH, hardness, and alkalinity range of most natural waters, were used. The characteristics of these waters were as follows:

| | D.O. | pH | Alkalinity | Hardness |
|---|---|---|---|---|
| Soft water | 9.0 | 7.4 | 30 | 40 |
| Hard water | 8.5 | 8.2 | 325 | 400 |

Fathead minnows (Pimephales promelas) were the test fish.

As the chemical was of extremely low solutility in water, two formulations were used in preparing test concentrations (1) a 0.01% acetone solution and (2) a 0.001% water suspension or emulsion prepared by mixing in a Waring Blender. The results of these tests are shown in Table 3. The data on which these results are based are included in the Appendix.

TABLE 3

TOXICITY OF HEXACHLOROCYCLOPENTADIENE

| Kind of Water | $TL_m$ (ppm compound) 24 hr. | 48 hr. | 96 hr. |
|---|---|---|---|
| **Acetone Solution** | | | |
| Soft | 0.115 | 0.110 | 0.104 |
| Hard | 0.093 | 0.078 | 0.078 |
| **Waring Blender Emulsion** | | | |
| Hard | 0.075 | 0.059 | 0.059 |

Hexachlorocyclopentadiene is highly toxic to fish and only slightly less toxic to fish than some of the most highly toxic insecticides, dieldrin and toxaphene. An estimated "safe" concentration for aquatic life would be somewhere in the range of from 5 to 10 parts per billion. Apparently the toxicity

was slightly greater in hard water which is in agreement with results previously determined for dieldrin.

Due to its highly toxic nature, this material should be carefully controlled to assure protection of aquatic life in receiving waters where aquatic life protection is desired.

APPENDIX TABLE 1 -BIOASSAY DATA

HOOKER ELECTROCHEMICAL COMPANY
NIAGARA FALLS, NEW YORK

TEST FISH - FATHEAD MINNOWS
DILUTION WATER - LAKE ERIE

| SOURCE AND METHOD OF COLLECTION OF SAMPLE | DATE OF COLLECTION | CONCENTRATION TESTED PERCENT | NUMBER OF TEST FISH | VOLUME OF TEST SAMPLE LITERS | TIME OF DEATH | | | PERCENT SURVIVAL | | | | CHEMICAL AND PHYSICAL DATA | | | | | | | REMARKS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | FIRST FISH | HALF OF FISH | ALL OF FISH | 24 HOURS | 48 HOURS | 72 HOURS | 96 HOURS | TIME | TEMP. °C | DISSOLVED OXYGEN ppm. | pH | TOTAL ALK ($CaCO_3$) ppm. | TOTAL ACIDITY ($CaCO_3$) ppm | VERSENATE HARDNESS ppm. | |
| Main Sewer | 1-20-56 | 100 | - | - | | | | | | | | Effluent | 23 | 6.2 | 7.0 | 0 | 740 | 1000 | Grayish cast gray suspended material-settles to clear solution. Sweetish odor. |
| (47th Street) | | | | | | | | | | | | | | | | | | | |
| 24 hr.composite | | 32 | 2 | 2 | 15 min. | 15 min. | 30 min. | 0 | 0 | 0 | 0 | Initial | 23 | - | 2.7 | - | - | - | |
| In Lab | | | | | | | | | | | | | 23 | - | - | - | - | - | |
| 4:00 PM 1/20 | | 18 | 10 | 20 | 30 min. | 30 min. | 1 hour | 0 | 0 | 0 | 0 | Initial | 23 | 7.6 | 3.1 | 0 | 102 | 290 | |
| Fish Added | | | | | | | | | | | | 1 hr. | 23 | 7.6 | 3.1 | 0 | 108 | 290 | Low pH |
| 2:00 PM 1/21 | | 10 | 10 | 20 | 4 hrs. | 5 hrs. | 5-21 hrs. | 0 | 0 | 0 | 0 | Initial | | 7.6 | 5.6 | 12 | 44 | 225 | |
| | | | | | | | | | | | | 21 hrs. | | 7.2 | 5.9 | 8 | 22 | 220 | |
| | | 5.6 | 10 | 20 | 5-21 hrs. | 5-21 hrs. | 5-21 hrs. | 0 | 0 | 0 | 0 | Initial | | 7.6 | 6.8 | 46 | 26 | 175 | |
| | | | | | | | | | | | | 21 hrs. | | 7.2 | 6.8 | 50 | 10 | 180 | |
| | | 3.2 | 10 | 20 | 5-21 hrs. | 5-21 hrs. | - | 50 | 20 | 10 | 10 | Initial | | 7.8 | 7.2 | 68 | 16 | 160 | |
| | | | | | | | | | | | | 96 hrs. | | 6.2 | 7.6 | 70 | 4 | 140 | |
| | | 1.8 | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 7.6 | 7.2 | 82 | 8 | 145 | |
| | | | | | | | | | | | | 96 hrs. | | 4.2 | 7.6 | 84 | 6 | 135 | |
| | | Control | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | 23 | 8.0 | 8.1 | 96 | 2 | 130 | |
| | | | | | | | | | | | | 96 hrs. | 23 | 5.0 | 7.6 | 100 | 6 | 125 | |
| | | TLm (Median Tolerance Limit - Per Cent Concentration) | | | | | | 3.2 | 2.6 | 2.5 | 2.5 | | | | | | | | |
| Main Sewer | 2-17-56 | 100 | - | - | | | | | | | | Effluent | 25 | 0.4 | 2.1 | 0 | 1450 | 550 | Gray color. Large amount of suspended material. Clear when settled. Sweetish solvent odor. |
| (47th Street) | | | | | | | | | | | | | | | | | | | |
| 24 hr.composite | | 32 | 2 | 2 | 15 min. | 15 min. | 15 min. | 0 | 0 | 0 | 0 | Initial | 25 | - | - | - | - | - | |
| In Lab | | | | | | | | | | | | 15 min. | 25 | 5.8 | 2.6 | - | - | - | |
| 11:00 AM 2/17 | | 18 | 2 | 2 | 15 min. | 15 min. | 15 min. | 0 | 0 | 0 | 0 | Initial | 25 | - | - | - | - | - | |
| Fish Added | | | | | | | | | | | | 15 min. | 25 | 6.6 | 2.9 | - | - | - | Low pH |
| 2:15 PM 2/17 | | 10 | 10 | 20 | 15 min. | 15 min. | 30 min. | 0 | 0 | 0 | 0 | Initial | | 6.8 | 3.4 | 0 | 104 | 180 | |
| | | | | | | | | | | | | 30 min. | | 7.0 | 3.4 | 0 | 104 | 180 | Low pH |
| | | 5.6 | 10 | 20 | 3-20 hrs. | 3-20 hrs. | 3-20 hrs. | 0 | 0 | 0 | 0 | Initial | | 7.0 | 5.8 | 14 | 38 | 145 | |
| | | | | | | | | | | | | 20 hrs. | | 6.4 | 6.2 | 12 | 14 | 145 | |
| | | 3.2 | 10 | 20 | 24 hrs. | - | - | 90 | 90 | 90 | 90 | Initial | | 7.4 | 6.5 | 50 | 26 | 135 | |
| | | | | | | | | | | | | 96 hrs. | | 5.0 | 7.3 | 54 | 4 | 140 | |
| | | 1.8 | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 7.4 | 7.0 | 70 | 14 | 130 | |
| | | | | | | | | | | | | 96 hrs. | | 4.8 | 7.3 | 74 | 4 | 135 | |
| | | Control | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | 25 | 7.6 | 8.1 | 100 | 2 | 125 | |
| | | | | | | | | | | | | 96 hrs. | 25 | 4.8 | 7.5 | 100 | 4 | 130 | |
| | | TLm (Median Tolerance Limit - Per Cent Concentration) | | | | | | 4.1 | 4.1 | 4.1 | 4.1 | | | | | | | | |
| Same as above | | 100 | - | - | | | | | | | | Effluent | 25 | 3.0 | 8.0 | 70 | 2 | 2200 | No additional precipitate when neutralized. |
| (Neutralized | | | | | | | | | | | | | | | | | | | |
| with lime) | | 10 | 5 | 10 | 2-20 hrs. | 2-20 hrs. | 2-20 hrs. | 0 | 0 | 0 | 0 | Initial | 25 | 7.2 | 8.0 | 96 | 2 | 340 | |
| Fish Added | | | | | | | | | | | | 20 hrs. | 25 | 4.4 | 7.6 | 96 | 8 | 330 | |
| 4:15 PM 2/28 | | 5.6 | 5 | 10 | 2-20 hrs. | 2-20 hrs. | 24-40 hrs. | 60 | 40 | 20 | 20 | Initial | | 7.4 | 8.1 | 96 | 2 | 240 | |
| | | | | | | | | | | | | 96 hrs. | | 5.6 | 7.5 | 100 | 4 | 250 | |
| | | 3.2 | 5 | 10 | | | | 100 | 100 | 100 | 100 | - | | - | - | - | - | - | |
| TLm (Median Tolerance Limit - Per Cent Concentration) | | | | | | | | 6.2 | 5.0 | 4.4 | 4.4 | | | | | | | | |

APPENDIX TABLE 2 -BIOASSAY DATA

HOOKER ELECTROCHEMICAL COMPANY
NIAGARA FALLS, NEW YORK

TEST FISH - FATHEAD MINNOWS
DILUTION WATER - LAKE ERIE

| SOURCE AND METHOD OF COLLECTION OF SAMPLE | DATE OF COLLECTION | CONCEN-TRATION TESTED PERCENT | NUMBER OF TEST FISH | VOLUME OF TEST SAMPLE LITERS | TIME OF DEATH | | | PERCENT SURVIVAL | | | | CHEMICAL AND PHYSICAL DATA | | | | | | | REMARKS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | FIRST FISH | HALF OF FISH | ALL OF FISH | 24 HOURS | 48 HOURS | 72 HOURS | 96 HOURS | TIME | TEMP. °C | DISSOLVED OXYGEN ppm. | p H | TOTAL ALK ($CaCO_3$) ppm. | TOTAL ACIDITY ($CaCO_3$) ppm | VERSENATE HARDNESS ppm. | |
| Main Sewer | 3-15-56 | 100 | - | - | | | | | | | | Effluent | 24 | 3.4 | 2.5 | 0 | 230 | 260 | Gray color. Turbid. Sour solvent odor. Low pH. |
| (47th Street) | | | | | | | | | | | | | | | | | | | |
| 24 hr.composite | | 32 | 5 | 10 | 4 hrs. | 5 hrs. | 6-20 hrs. | 0 | 0 | 0 | 0 | Initial | 24 | 7.0 | 4.5 | 0 | 52 | 185 | |
| In Lab | | | | | | | | | | | | 20 hrs. | 24 | 7.0 | 4.6 | 4 | 24 | 165 | |
| 6:00 PM 3/15 | | 18 | 5 | 10 | 29-41 hrs | - | - | 80 | 80 | 80 | 80 | Initial | | 7.2 | 6.5 | 40 | 30 | 150 | |
| Fish Added | | | | | | | | | | | | 96 hrs. | | 5.4 | 7.1 | 42 | 6 | 160 | |
| 3:30 PM 3/17 | | 10 | 5 | 10 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 7.4 | 7.1 | 60 | 16 | 140 | |
| | | | | | | | | | | | | 96 hrs. | | 6.0 | 7.2 | 66 | 6 | 160 | |
| | | Control | 5 | 10 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 7.8 | 8.3 | 92 | 2 | 115 | |
| | | | | | | | | | | | | 96 hrs. | | 6.2 | 7.6 | 100 | 6 | 130 | |
| | | TLm (Median Tolerance Limit - Per Cent Concentration) | | | | | | 22 | 22 | 22 | 22 | | | | | | | | |
| | | | | | | | | | | | | | | | | | | | |
| Same as above | | 100 | - | - | | | | | | | | Effluent | 24 | 7.0 | 7.5 | 24 | 0 | 500 | Fairly heavy light gray precipitate. |
| Neutralized | | | | | | | | | | | | 96 hrs. | | | | | | | |
| with lime | | 56 | 2 | 2 | - | - | - | 100 | 100 | 100 | | Initial | 24 | - | - | - | - | - | |
| Fish Added | | | | | | | | | | | | 96 hrs. | 24 | 5.8 | 7.3 | 50 | 8 | 365 | |
| 2:00 PM 3/19 | | 32 | 5 | 10 | - | - | - | 100 | 100 | 100 | | Initial | | 7.4 | 7.9 | 74 | 4 | 240 | |
| | | | | | | | | | | | | 96 hrs. | | 4.8 | 7.5 | 74 | 6 | 350 | |
| | | TLm (Median Tolerance Limit - Per Cent Concentration) | | | | | | >56 | >56 | >56 | >56 | | | | | | | | Did not test any higher concentration. |
| | | | | | | | | | | | | | | | | | | | |
| Diversion sewer | 2-21-56 | 100 | 5 | 10 | - | - | - | 100 | 100 | 100 | 100 | Initial | 25 | 8.6 | 8.6 | 104 | 0 | 160 | Grayish tan color. Slightly turbid. Very faint solvent odor. |
| 8 hr.composite | | | | | | | | | | | | 96 hrs. | 25 | 4.4 | 7.7 | 112 | 4 | 165 | |
| In Lab | | 56 | 2 | 2 | - | - | - | 100 | 100 | 100 | 100 | - | | - | - | - | - | - | |
| 6:00 PM 2/21 | | | | | | | | | | | | | | - | - | - | - | - | |
| Fish Added | | 32 | 2 | 2 | - | - | - | 100 | 100 | 100 | 100 | - | | - | - | - | - | - | |
| 7:00 PM 2/21 | | | | | | | | | | | | | | - | - | - | - | - | |
| | | Control | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 7.8 | 8.1 | 102 | 2 | 130 | |
| | | | | | | | | | | | | 96 hrs. | | 5.0 | 7.6 | 102 | 4 | 130 | |
| | | | | | | | | No Fish Mortality | | | | | | | | | | | |

APPENDIX TABLE 3 -BIOASSAY DATA

HOOKER ELECTROCHEMICAL COMPANY
NIAGARA FALLS, NEW YORK

TEST FISH - FATHEAD MINNOWS
DILUTION WATER - LAKE ERIE

| SOURCE AND METHOD OF COLLECTION OF SAMPLE | DATE OF COLLECTION | CONCENTRATION TESTED PERCENT | NUMBER OF TEST FISH | VOLUME OF TEST SAMPLE LITERS | TIME OF DEATH | | | PERCENT SURVIVAL | | | | CHEMICAL AND PHYSICAL DATA | | | | | | | REMARKS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | FIRST FISH | HALF OF FISH | ALL OF FISH | 24 HOURS | 48 HOURS | 72 HOURS | 96 HOURS | TIME | TEMP. °C | DISSOLVED OXYGEN ppm. | p H | TOTAL ALK ($CaCO_3$) ppm. | TOTAL ACIDITY ($CaCO_3$) ppm | VERSENATE HARDNESS ppm. | |
| West End | 2-21-56 | 100 | - | - | | | | | | | | Effluent | 25 | 9.2 | 2.1 | 0 | 1020 | 175 | Light grey color. Some suspended material. |
| Sewer | | | | | | | | | | | | | | | | | | | |
| 8 hr.composite | | 18 | 2 | 2 | 30 min. | 30 min. | 30 min. | 0 | 0 | 0 | 0 | Initial | | - | - | - | - | - | Sweetish solvent odor. |
| In Lab | | | | | | | | | | | | | | | | | | | |
| 2:00 PM 6/21 | | 10 | 5 | 10 | 30 min. | 1 hr. | 1 hr. | 0 | 0 | 0 | 0 | Initial | 25 | 8.0 | 3.9 | 0 | 76 | 135 | |
| Fish Added | | | | | | | | | | | | 1 hr. | 25 | 7.0 | 4.2 | 0 | 68 | 130 | Low pH |
| 2:00 PM 2/22 | | 5.6 | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 8.0 | 6.2 | 40 | 38 | 135 | |
| | | | | | | | | | | | | 96 hrs. | | 4.4 | 7.0 | 46 | 6 | 130 | |
| | | 3.2 | 10 | 20 | 27-43 | - | - | 90 | 90 | 90 | 90 | Initial | | 8.0 | 6.6 | 66 | 24 | 130 | |
| | | | | | | | | | | | | 96 hrs. | | 5.4 | 7.4 | 66 | 4 | 130 | |
| | | Control | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 7.8 | 8.1 | 102 | 2 | 130 | |
| | | | | | | | | | | | | 96 hrs. | | 5.0 | 7.6 | 102 | 4 | 130 | |
| | | TLm (Median Tolerance Limit - Per Cent Concentration) | | | | | | 7.5 | 7.5 | 7.5 | 7.5 | | | | | | | | |
| Same as above | | 100 | - | - | | | | | | | | Effluent | 25 | 9.0 | 7.2 | 64 | 12 | 1200 | No precipitate. |
| Neutralized | | | | | | | | | | | | | | | | | | | |
| with lime | | 18 | 5 | 10 | 2-19 hrs. | 24 hrs. | - | 40 | 20 | 20 | 20 | Initial | 25 | 8.0 | 7.7 | 94 | 4 | 315 | |
| Fish Added | | | | | | | | | | | | 96 hrs. | 25 | 5.0 | 7.5 | 100 | 4 | 330 | |
| 2:30 PM 2/22 | | 10 | 5 | 10 | 2-19 hrs. | - | - | 80 | 80 | 80 | 80 | Initial | | 7.8 | 7.9 | 98 | 2 | 225 | |
| | | | | | | | | | | | | 96 hrs. | | 3.6 | 7.5 | 100 | 4 | 250 | |
| | | TLm (Median Tolerance Limit - Per Cent Concentration ) | | | | | | 16 | 13 | 13 | 13 | | | | | | | | |
| Plant #4 | 3-13-56 | 100 | - | - | | | | | | | | Effluent | 24 | 0.2 | 1.8 | 0 | 520 | 300 | Greyish-tan color. Slightly turbid. Sweetish solvent odor. |
| (North Street) | | | | | | | | | | | | | | | | | | | |
| 8 hr. composite | | 18 | 2 | 2 | 15 min. | 15 min. | 15 min. | 0 | 0 | 0 | 0 | Initial | 24 | - | - | - | - | - | |
| In Lab | | | | | | | | | | | | 15 min. | 24 | 7.0 | 2.5 | - | - | - | Low pH |
| 4:00 PM 3/13 | | 10 | 5 | 10 | 1 hr. | 2 hrs. | 2 hrs. | 0 | 0 | 0 | 0 | Initial | | 6.2 | 3.2 | 0 | 114 | 150 | |
| Fish Added | | | | | | | | | | | | 2 hrs. | | 5.2 | 3.1 | 0 | 104 | 140 | Low pH |
| 3:15 PM 3/17 | | 5.6 | 5 | 10 | 5 hrs. | 6 hrs. | 7 hrs. | 0 | 0 | 0 | 0 | Initial | | 6.4 | 5.5 | 14 | 48 | 140 | |
| | | | | | | | | | | | | 7 hrs. | | 6.4 | 5.8 | 16 | 34 | 140 | |
| | | 3.2 | 5 | 10 | 7-20 hrs | 7-20 hrs | 24 hrs. | 0 | 0 | 0 | 0 | Initial | | 6.8 | 6.4 | 48 | 24 | 130 | |
| | | | | | | | | | | | | 24 hrs. | | 7.0 | 7.2 | 46 | 8 | 145 | |
| | | 1.8 | 5 | 10 | 24 hrs. | 32-48 hrs | - | 80 | 40 | 40 | 40 | Initial | | 7.0 | 7.1 | 66 | 24 | 125 | |
| | | | | | | | | | | | | 96 hrs. | | 5.8 | 7.4 | 70 | 8 | 140 | |
| | | 1.0 | 5 | 10 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 9.4 | 6.6 | 68 | 14 | 120 | |
| | | | | | | | | | | | | 96 hrs. | | 6.4 | 7.7 | 78 | 6 | 125 | |
| | | Control | 5 | 10 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 7.8 | 8.3 | 92 | 2 | 115 | |
| | | | | | | | | | | | | 96 hrs. | | 6.2 | 7.6 | 100 | 6 | 130 | |
| | | TLm (Median Tolerance Limit - Per Cent Concentration) | | | | | | 2.2 | 1.5 | 1.5 | 1.5 | | | | | | | | |

APPENDIX TABLE 4 -BIOASSAY DATA

HOOKER ELECTROCHEMICAL COMPANY
NIAGARA FALLS, NEW YORK

TEST FISH - FATHEAD MINNOWS
DILUTION WATER - HARD (HATCHERY-LIMESTONE SPRING)

| SOURCE AND METHOD OF COLLECTION OF SAMPLE | DATE OF COLLECTION | CONCENTRATION TESTED ppm | NUMBER OF TEST FISH | VOLUME OF TEST SAMPLE LITERS | TIME OF DEATH | | | PERCENT SURVIVAL | | | | CHEMICAL AND PHYSICAL DATA | | | | | | | REMARKS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | FIRST FISH | HALF OF FISH | ALL OF FISH | 24 HOURS | 48 HOURS | 72 HOURS | 96 HOURS | TIME | TEMP. °C | DISSOLVED OXYGEN ppm. | p H | TOTAL ALK ($CaCO_3$) ppm. | TOTAL ACIDITY ($CaCO_3$) ppm | VERSENATE HARDNESS ppm. | |
| Hexachlorocyclopentadiene | | | | | | | | | | | | | | | | | | | |
| Pure chemical | 2-7-56 | 16 | 2 | 2 | 15 min. | 15 min. | 30 min. | 0 | 0 | 0 | 0 | - | 22 | - | - | - | - | - | |
| 0.01% Acetone | | | | | | | | | | | | | | | | | | | |
| solution used | | 3.2 | 2 | 2 | 30 min. | 30 min. | 45 min. | 0 | 0 | 0 | 0 | - | | - | - | - | - | - | |
| in preparing | | | | | | | | | | | | | | | | | | | |
| dilutions | | 1.6 | 2 | 2 | 2 hrs. | 2 hrs. | 3 hrs. | 0 | 0 | 0 | 0 | - | | - | - | - | - | - | |
| | | .56 | 2 | 2 | 4 hrs. | 4 hrs. | 5 hrs. | 0 | 0 | 0 | 0 | - | | - | - | - | - | - | |
| | | .32 | 10 | 20 | 3-19 hrs. | 3-19 hrs. | 3-19 hrs. | 0 | 0 | 0 | 0 | Initial | 22 | 9.0 | 8.2 | 322 | 2 | 380 | |
| | | | | | | | | | | | | 19 hrs. | 22 | 7.2 | 8.2 | 320 | 2 | 378 | |
| | | .18 | 10 | 20 | 3-19 hrs. | 3-19 hrs. | 3-19 hrs. | 0 | 0 | 0 | 0 | Initial | | 8.8 | 8.2 | 342 | 1 | 408 | |
| | | | | | | | | | | | | 19 hrs. | | 7.2 | 8.2 | 338 | 2 | 394 | |
| | | .10 | 10 | 20 | 3-19 hrs. | 3-24 hrs. | 27-43 hrs. | 40 | 0 | 0 | 0 | Initial | | 8.2 | 8.3 | 340 | 2 | 404 | |
| | | | | | | | | | | | | 43 hrs. | | 5.6 | 8.2 | 340 | 2 | 402 | |
| | | .075 | 10 | 20 | 3-19 hrs. | - | - | 80 | 60 | 60 | 60 | Initial | | 8.0 | 8.2 | 340 | 2 | 400 | |
| | | | | | | | | | | | | 96 hrs. | | 3.8 | 8.2 | 300 | 2 | 366 | |
| | | .056 | 10 | 20 | 72 hrs. | - | - | 100 | 100 | 90 | 80 | Initial | | 8.6 | 8.2 | 332 | 2 | 392 | |
| | | | | | | | | | | | | 96 hrs. | | 6.0 | 8.2 | 340 | 2 | 404 | |
| | | Control | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 8.4 | 8.4 | 324 | 0 | 380 | |
| | | | | | | | | | | | | 96 hrs. | | 4.6 | 8.2 | 288 | 2 | 348 | |
| | | TLm (Median Tolerance Limit - ppm) | | | | | | .093 | .078 | .078 | .078 | | | | | | | | |
| Hexachlorocyclopentadiene | | | | | | | | | | | | | | | | | | | |
| Pure chemical | 3-5-56 | .18 | 10 | 20 | 3 hrs. | 5 hrs. | 7 hrs. | 0 | 0 | 0 | 0 | Initial | 22 | 7.8 | 8.2 | 310 | 2 | 460 | |
| Emulsion | | | | | | | | | | | | 7 hrs. | 22 | 8.0 | 8.0 | 230 | 2 | 360 | |
| (Prepared in | | .10 | 10 | 20 | 7-23 hrs. | 7-23 hrs. | 7-23 hrs. | 0 | 0 | 0 | 0 | Initial | | 8.0 | 8.2 | 314 | 2 | 440 | |
| Waring blender) | | | | | | | | | | | | 23 hrs. | | 7.6 | 8.0 | 278 | 2 | 352 | |
| .001% concentration | | .075 | 10 | 20 | 7-23 hrs. | 24 hrs. | 30 hrs. | 50 | 0 | 0 | 0 | Initial | | 8.4 | 8.2 | 300 | 2 | 380 | |
| used for dilutions | | | | | | | | | | | | 30 hrs. | | 8.2 | 8.0 | 270 | 2 | 324 | |
| | | .056 | 10 | 20 | 7-23 hrs. | - | - | 80 | 60 | 60 | 60 | Initial | | 8.2 | 8.2 | 326 | 2 | 370 | |
| | | | | | | | | | | | | 96 hrs. | | 7.2 | 8.2 | 302 | 2 | 364 | |
| | | .032 | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 8.6 | 8.2 | 310 | 2 | 380 | |
| | | | | | | | | | | | | 96 hrs. | | 6.6 | 8.2 | 274 | 2 | 328 | |
| | | Control | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | Initial | | 8.6 | 8.2 | 328 | 2 | 430 | |
| | | | | | | | | | | | | 96 hrs. | | 6.6 | 8.2 | 284 | 2 | 350 | |
| | | TLm (Median Tolerance Limit - ppm) | | | | | | .078 | .059 | .059 | .059 | | | | | | | | |

APPENDIX TABLE 5 -BIOASSAY DATA

HOOKER ELECTROCHEMICAL COMPANY
NIAGARA FALLS, NEW YORK

TEST FISH - FATHEAD MINNOWS
DILUTION WATER - SOFT (5% HATCHERY)

| SOURCE AND METHOD OF COLLECTION OF SAMPLE | DATE OF COLLECTION | CONCENTRATION TESTED ~~[illegible]~~ PPM | NUMBER OF TEST FISH | VOLUME OF TEST SAMPLE LITERS | TIME OF DEATH | | | PERCENT SURVIVAL | | | | CHEMICAL AND PHYSICAL DATA | | | | | | | REMARKS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | FIRST FISH | HALF OF FISH | ALL OF FISH | 24 HOURS | 48 HOURS | 72 HOURS | 96 HOURS | TIME | TEMP. °C | DISSOLVED OXYGEN ppm. | p H | TOTAL ALK ($CaCO_3$) ppm. | TOTAL ACIDITY ($CaCO_3$) ppm | VERSENATE HARDNESS ppm. | |
| Hexachlorocyclopentadiene | | | | | | | | | | | | | | | | | | | |
| Pure Chemical | 2-7-56 | 18 | 2 | 2 | 15 min. | 15 min. | 30 min. | 0 | 0 | 0 | 0 | - | 22 | - | - | - | - | - | |
| 0.01% Acetone solution used | | 3.2 | 2 | 2 | 30 min. | 30 min. | 45 min. | 0 | 0 | 0 | 0 | - | | - | - | - | - | - | |
| in preparing | | | | | | | | | | | | | | | | | | | |
| dilutions | | 1.8 | 2 | 2 | 2 hrs. | 2 hrs. | 3 hrs. | 0 | 0 | 0 | 0 | - | | - | - | - | - | - | |
| | | .56 | 2 | 2 | 6 hrs. | 6-22 hrs. | 6-22 hrs. | 0 | 0 | 0 | 0 | - | | - | - | - | - | - | |
| | | .32 | 10 | 20 | 3-19 hrs. | 3-19 hrs. | 3-19 hrs. | 0 | 0 | 0 | 0 | Initial | 22 | 6.8 | 7.3 | 42 | 2 | 30 | |
| | | | | | | | | | | | | 19 hrs. | 22 | 7.6 | 6.9 | 28 | 3 | 30 | |
| | | .18 | 10 | 20 | 3-19 hrs. | 3-19 hrs. | 3-19 hrs. | 0 | 0 | 0 | 0 | Initial | | 8.6 | 7.3 | 24 | 1 | 24 | |
| | | .135 | 10 | 20 | 3-19 hrs. | 3-19 hrs. | 3-19 hrs. | 0 | 0 | 0 | 0 | 19 hrs. | | 7.2 | 6.9 | 16 | 4 | 28 | |
| | | | | | | | | | | | | Initial | | 8.6 | 7.1 | 26 | 3 | 26 | |
| | | .10 | 10 | 20 | 19-24 hrs | - | - | 90 | 80 | 70 | 60 | 19 hrs. | | 7.6 | 7.0 | 30 | - | 30 | |
| | | | | | | | | | | | | Initial | | 8.0 | 7.3 | 26 | 3 | 26 | |
| | | .056 | 10 | 20 | 72 hrs. | - | - | 100 | 100 | 90 | 90 | 96 hrs. | | 6.4 | 7.1 | 30 | 2 | 30 | |
| | | | | | | | | | | | | Initial | | 8.6 | 7.2 | 32 | 1 | 40 | |
| | | Control | 10 | 20 | - | - | - | 100 | 100 | 100 | 100 | 96 hrs. | | 5.0 | 7.1 | 44 | 3 | 42 | |
| | | | | | | | | | | | | Initial | | 9.0 | 7.3 | 38 | 2 | 46 | |
| | | Tlm (Median Tolerance Limit - ppm) | | | | | | 0.115 | 0.11 | 0.107 | 0.104 | 96 hrs. | | 4.8 | 7.1 | 42 | 3 | 48 | |